HEREDITY AND THE GERM-CELLS.

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THE CARTWRIGHT LECTURES FOR 1892, III.

(*Continued from Page 567, Vol. XXVI.*)

According to the general law[1] the germ-cell is considered as matter potentially alive and having within itself the tendency to assume a definite living form in course of individual development. The nucleus must be extraordinarily complex, for it contains within itself not only the tendencies of the present type, but of past types far distant. The supposition of a vast number of germs of structure is required by the phenomena of heredity; Nägeli has demonstrated that even in so minute a space as $\frac{1}{1000}$ cub. millimetre, 400,000,000 micellæ must be present.

The study of heredity will ultimately centre around the structure and functions of the germ-cells. The precise researches of Galton show that the external facts of heredity, questions of averages and of probabilities, of paternal and maternal contributions to the offspring, are capable of being reduced to an exact science in which mathematical calculations will enable us to forecast the characteristics of the coming generation.

There will still remain, however, a large residuum of facts which will present themselves to a mathematician like Galton as chance or inexact, such as the physiological conditions of reversion; the causes of prepotency, by which the maternal or the paternal characteristics prevail in parts or in the entire structure of the offspring; the material basis of latent heritage upon which reversion depends, and which compels us to hypothecate either an unused hereditary substance or a return to an older disposition of the forces in this substance; the nature and determination of sex. These apparently chance

[1]See Huxley, Article Evolution, Enc. Britannica, p. 746.

phenomena must also be due to certain fixed laws, and by far the most promising routes to discovery have already been taken by Van Beneden, the Hertwig brothers, Boveri, Maupas, and others.

They have attacked the problem of the relation of the germ-cells to heredity on every side, and by the most ingenious and novel methods, which are familiar enough in various branches of gross anatomical and physiological research, but seem almost out of the limits of application to minute microscopic objects. For example, the Hertwig brothers have ascertained the influences of various solutions of morphine and other drugs, of the alcohols, and of various degrees of temperature upon the ovum and spermatozoon during the conjugation

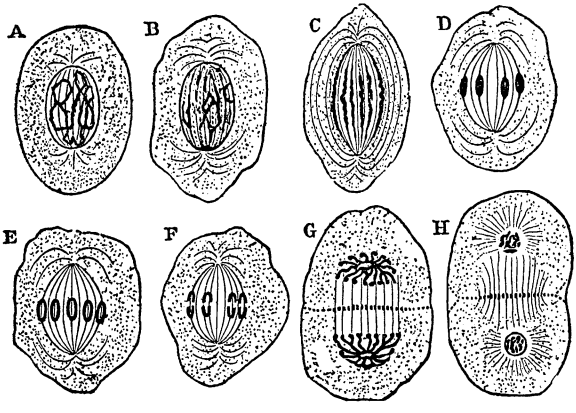

FIG. 7.—TYPICAL CELL DIVISION, SHOWING THE DISTRIBUTION OF CHROMATIN. (From Parker, after Carnoy) A-C, Arrangement of the chromatin in threads; D-E, Formation of the chromatin rods and loops; F, Splitting of the loops; G-H, Retraction of the chromatin into the two daughter-cells.

period, with results which are highly suggestive of the causes of congenital malformations, anomalies, and double births. The Hertwigs and Boveri have succeeded in robbing ova of their nuclei, and watching the results of the subsequent entrance of spermatozoa. In order to further test the relations of the nucleus to the remainder of the cell, Verworn has experimented along the same line with extirpations of every kind from the single cells of Infusoria. Of equal novelty are

the recent studies of Maupas upon the multiplication and conjugation of the Infusoria, giving us a host of new ideas as to the cycle of life, the meaning of sex, and the origin of the sexual relation.

In all this research and in the future outlook there are two main questions:

1. *What is the hereditary substance?* What is the material basis of heredity, which spreads from the fertilized ovum to every cell in the body, conveying its ancestral characteristics? Is there any substance corresponding to the hypothetical idioplasm of Nägeli?

2. *What are its regulating and distributing forces?* How is the hereditary substance divided and distributed? How far is it active or passive?

I may say at the outset that the idioplasm of Nägeli, a purely ideal element of protoplasm which he conceived of as permeating all the tissues of the body as the vehicle of heredity, has been apparently materialized in the *chromatin* or highly coloring materials in the centre of the nucleus. This rests upon the demonstration by Van Beneden and others that chromatin is found not only in all active cells, but is a conspicuous element in both the ovum and spermatozoon during all the phenomena attending conjugation.

Secondly, that while the chromatin is apparently passive, it is played upon by forces resident in the clear surrounding protoplasm of the nucleus, but chiefly by the extra nuclear *archoplasm*, which seems to constitute the dynamic and mechanical factor in each cell. This, unlike the chromatin, only comes into view when there is unusual activity, as during cell-division, and is not evident (with our present histological technique, at least), when the cell is arrested by reagents in any of the ordinary stages of metabolism.

The Distribution of Hereditary Substance.—I may first review some of the well-known phenomena attending the distribution of the chromatin substance to the tissues.

I have borrowed from Parker figures by Carnoy to illustrate the resting and active stages of the cell, and from Watase, a

Japanese student of Clark University, figures representing the high differentiation of the cell-contents during division (figs. 8, 9). They bring out the active and passive elements of the typical cell.

The phenomena of karyokinesis which attend the division and distribution of the hereditary substance throughout the whole course of embryonic and adult development are well illustrated in Carnoy's figures (fig. 7). First we have the qui-

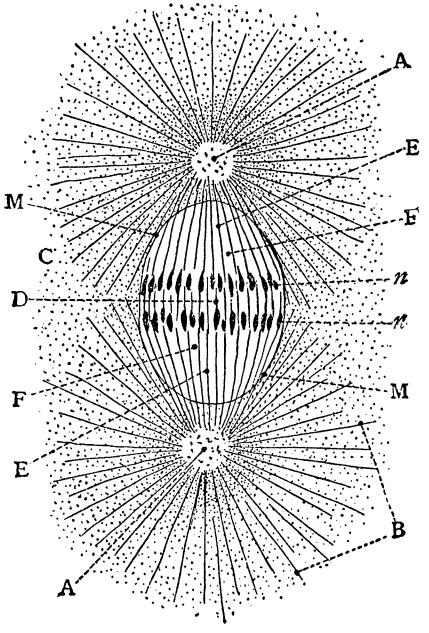

FIG. 8.—BEFORE DIVISION. DIFFERENTIATION OF THE CYTOPLASM AND NUCLEUS DURING CELL DIVISION OF A SQUID EMBRYO, LOLIGO. (After Watase.) M, The nuclear membrane; F, Achromatin or nucleoplasm; C, Cytoplasm, or protoplasm outside of the nucleus; A-A, The two centrosomes of archoplasm, B, Extra-nuclear archoplasmic filaments; E, Intra-nuclear archoplasmic filaments attached to *n*, *n'*, the chromatin rods.

escent period, in which the chromatin presents the appearance of a coiled, tangled thread; surrounding this is the clear nucleoplasm (or achromatin) bounded by the nuclear membrane; the extra-nuclear substance, or cytoplasm, is apparently undifferentiated. As soon as cell division sets in, however, radiating lines are seen in the cytoplasm above and below the

nucleus, these are called the archoplasmic filaments by Boveri, since they proceed from what is now believed to be the dynamic element, the archoplasm (fig. 8). As the activity becomes more intense the filaments are seen to diverge from a centre—the archoplasmic *centrosome*—which lies just without the nucleus at either pole; this radial display of cell-forces suggested the term "asters" to Fol, and "spheres attractive" to Van Beneden. The behavior of the chromatin, or hereditary substance, under these archoplasmic forces, is beautifully shown in Carnoy's diagrams (fig. 7). First, the nuclear wall

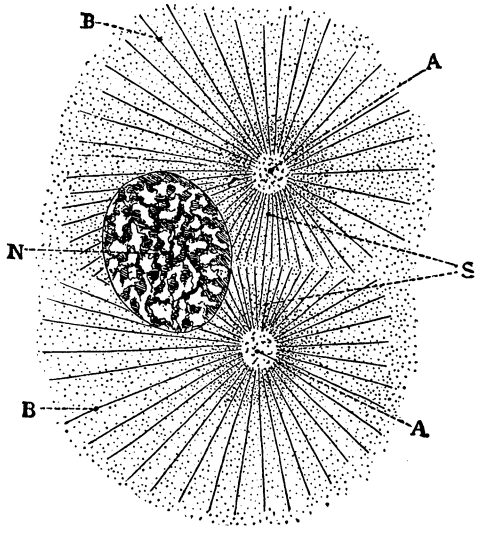

FIG. 9.—AFTER DIVISION, INTERIOR OF A DAUGHTER-CELL IN THE SQUID. (After Watase.) Division has just taken place and the daughter-nucleus, N, shows the chromatin coil. The daughter centrosome is just forming two new centrosomes, A-A, by direct division.

breaks up, then the chromatin coil unfolds into lines of vertical striation which become thread-like, hence the term mitosis, and then more compact, until finally a number of distinct vertical rods, chromatin rods, or *chromasomes* are formed.

A remarkable and significant fact may be noted here, that the number of chromasomes varies in the cells of different species, and even in the cells of different varieties (as in the thread-worm of the horse—*Ascaris megalocephala*), but is con-

stant in all the cells of the same variety through all stages; thus the same number of chromasomes appears in the first segmentation of the fertilized ovum as in the subsequent cell division in the tissues.

Carnoy next indicates the vertical splitting of each rod into a loop or link preceding the horizontal splitting; thus we may conceive of a thorough redistribution of the chromatin before it passes into the daughter-cells. The split loops are each retracted toward a centrosome, suggesting to some authors a contractile power in the archoplasmic filaments; each chromasome being apparently withdrawn by a single filament. But as the chromasomes separate, the filaments also appear between them, and are variously termed "interzonal," "verbindungs Fäden," "filaments réunissant;" there is, therefore, some difference of opinion as to what the mechanics of the chromasome divisions really are. The chromatin is now retracted into two coiled threads, each the centre of the daughter-nucleus with a single centrosome beside it. But as the line of cleavage is drawn between the two cells (fig. 9), the single centrosome in each cell divides so that each daughter-cell is now complete with its chromatin coil and two archoplasmic centrosomes. This process has been beautifully described by Watase.[2]

It thus appears that both the chromatin and archoplasm are permanent elements of the cell, such as we formerly considered the nucleus; the apparently passive chromatin is divided with great precision by the active archoplasm, then the archoplasm simply splits in two to resume the cleavage function.

Fertilization—The Union of Hereditary Substances.—Before looking at the host of questions which fertilization suggests, let us review a few of the well-known phenomena preparatory to the union of the germ-cells, in order to give greater emphasis to the importance of recent discoveries.

First, the ovum is a single cell, the typical structure of which, with its nucleus and cytoplasm, is generally obscured

[2]See Marine Biological Laboratory Lectures, 1889. Boston: Ginn & Co.

by a quantity of food-material, surrounded by a rather dense cell-wall. The ovum is said to be ripened or "matured" for the reception of the spermatozoon, by the extrusion of two small "polar bodies," containing both chromatin and hyaline protoplasm, and separating off by karyokinetic division. After maturation is complete, a single spermatozoon normally penetrates; then a reaction immediately sets in in the cell-wall of the ovum, which prevents other spermatozoa from entering. The head of the spermatozoon and the nucleus of the ovum now fuse together to form a single nucleus, which obviously contains the hereditary substance of two individuals. This is the starting point of the segmentation or distribution process above described, and it follows that the fertilized ovum at this stage must contain its typical complement of chromatin, archoplasm, etc., for the whole course of growth to the adult.

How shall we connect these phenomena of fertilization with the facts of heredity? The most suggestive enigma in connection with the fertilization process has been *the meaning of the two polar bodies*, especially since Van Beneden demonstrated that they contained chromatin. For twenty-five years speculation has been rife as to why the ovum should extrude a portion of its substance in two small cells; why not in one cell? Why not in a larger number? Thanks to the intense curiosity which these polar bodies have aroused, and to the great variety of explanations which have been offered for them, we have arrived to-day at a solution which links the higher animals with the lower, breaks down the supposed barrier between the sexes, and accords with the main external facts of heredity.

It seems to me best to disregard the order of discovery, and to state the facts in the most direct way. First, a few words as to the speculations upon the meaning of the polar bodies.

The early views of fertilization[3] were naturally based upon the apparent significance of this process in the human species, in which the sexes are sharply distinguished from each other in their entire structure, and the reproductive cells are also

[3]See also the introduction of Weismann's last essay, Amphimixis.

widely differentiated in form, the ovum large and passive, the spermatozoon small and active. The readiest induction was to regard these elements as representing distinct physiological principles, corresponding to the essential sexual characteristics—in short, as male and female cells, the former vitalizing and rejuvenating the latter. Thus one of the earliest definite "polar-body" theories was that the ovum was hermaphrodite, containing both male and female principles, and that it was necessary to get rid of the male substance before the spermatozoon could enter.

As Von Siebold and Leuckart had demonstrated that some ova reproduce parthenogenetically, that is without fertilization by spermatozoa, Weismann turned to such forms for the solution of this problem, and was surprised to find that parthenogetic ova only extrude one polar body; this led him to attach one meaning to the first polar body and another meaning to the second, which he viewed as designed to reduce the hereditary substance in the ovum without regard to sex. Thus both this and the older theory conveyed alike the idea of *reduction*, but with an entirely different supposition as to the nature of the material reduced or eliminated.

Maupas on Conjugation among the Infusoria.[4]—Among the newer researches which throw light upon this old problem those of Maupas are certainly the most brilliant. After a most exact and arduous research, extending over several years, he collected his results in two memoirs, published in 1889 and 1890.

His experiments were first directed upon the laws of direct multiplication by fission, which revealed a complete cycle of life in the single-celled Infusoria and showed that after a long period this mode of reproduction becomes less vigorous, then declines, and finally ceases altogether unless the stock is rejuvenated by conjugation of individuals from different broods. In other words, these broods of minute organisms grow old and die unless they are enabled to fertilize each other by an

[4]Sur la multiplication des Infusoires Ciliés, Archiv de Zoologie expérimentale, Sér. 3, vol. vi, pp. 165–273; Le Rejeunissement Karyogamique chez les Ciliés, vol. vii, pp. 149–517. See also Hartog, Quart. Journ. Microscop. Science, December, 1891.

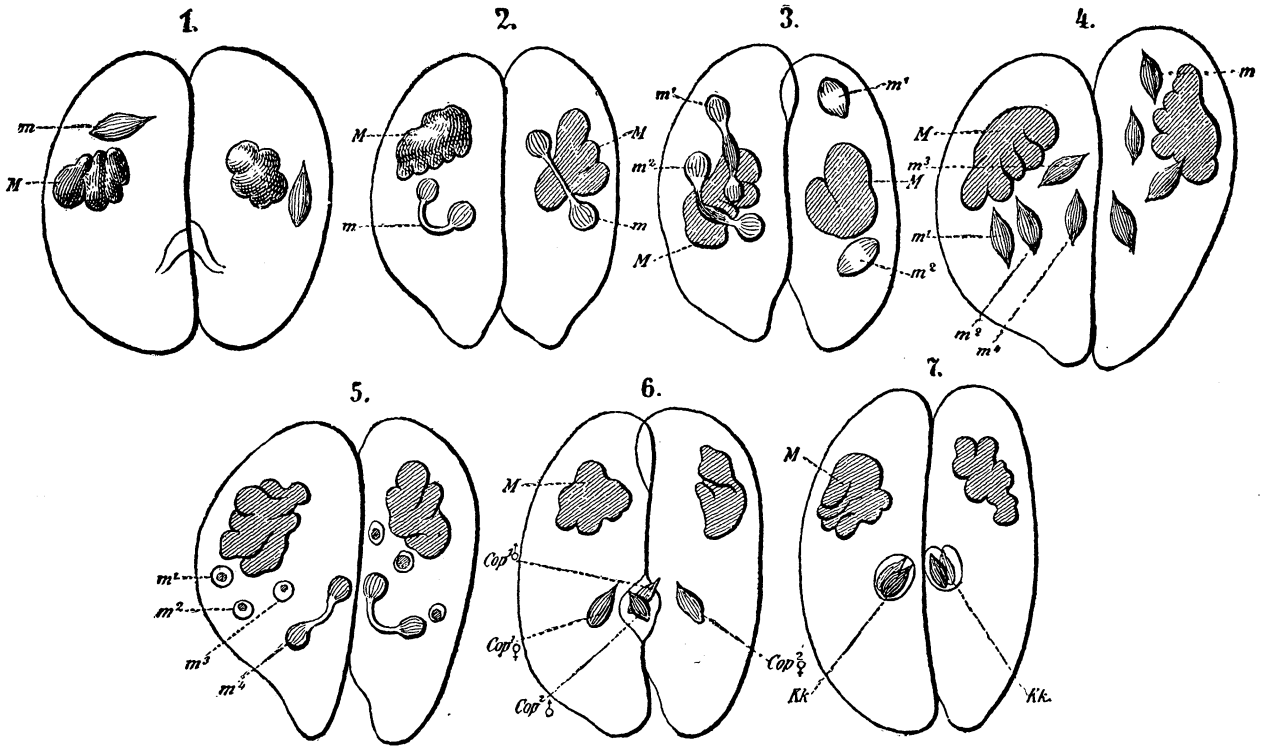

FIG. 10.—THE CONJUGATION OF INFUSORIA. (From Weismann, after Maupas.) 1, Two Infusoria copulating; *M*, meganucleus; *m*, micronucleus; 2-5, Successive divisions of micronuclei; 6, The migration of one of the persisting micronuclei from each infusorian into the other; 7, Union of the interchanged micronuclei.

exchange of hereditary substance altogether analogous to that observed in the higher multicellular organisms.

The cultures were made in a drop of water upon a slide, and feeding was adapted either to the herbivorous or carnivorous habits of the species. Under these conditions it was found that the rate of fission or direct multiplication varied directly with the temperature and food, rising in some species (*Glaucoma scintillans*) to five bipartitions daily. With the optimum of conditions this rate, if sustained for thirty-eight days, would produce from a single individual a mass of protoplasm equivalent to the volume of the sun. This rate is, however, found to be steady for a time, and then the offspring decline into "senescence," in which they appear at times only one-fourth the original size, with reduced buccal wreaths and degenerate nuclear apparatus. This is reached sooner in some species than in others; *Stylonichia pustulata* survives three hundred and sixteen generations or fissions, while *Leucophrys patula* persists to six hundred and sixty generations. Finally, even under the most favorable condition of environment, death ensues.

Not so where conjugation is brought about by mingling the offspring of different broods in the same fluid, as in the natural state. Maupas soon discovered that exhaustion of food would induce conjugation between members of mixed broods. He thus could watch every feature of the conjugation process, and determine all the phases in the cycle of life. These differed, as in the longevity of the species. In *Stylonichia*, for example, "immaturity" extended over the first one hundred bipartitions; "puberty," or the earliest phase favorable to conjugation, set in with the one hundred and thirtieth bipartition; "eugamy," or the most favorable conjugation phase, extended to the one hundred and seventieth; then "senescence" set in, characterized by a sexual hyperæsthesia in which conjugation was void of result or rejuvenescence, owing apparently to the destruction of the essential nuclear apparatus.

Conjugation begins with the approach of two individuals, and adhesion by their oral surfaces. There is no fusion, but an immediate transformation in the cell contents of each indi-

vidual sets in, concluding with an interchange of nuclear substance. In each cell Maupas distinguishes between the (*M*) *meganucleus* (fig. 10, the macronucleus, nucleus, endoplast of authors), which presides over nutrition and growth and divides by constriction, and the (*m*) *micronucleus* (paranucleus, nucleolus, of authors), which presides over the preservation of the species. The latter contains chromatin; it is the seat of rejuvenescence, the basis of heredity, it divides by mitosis, showing all the typical stages of karyokinesis excepting the loss of the cell membrane.

The transformation in each of these copulating cells first affects the centres of hereditary substance, viz., the micronuclei; they divide three times; thus the micronuclear substance is reduced to one-fourth of its original bulk. It is contained in two surviving micronuclei (the others being absorbed or eliminated), one of which migrates into the adjoining cell; the other remains stationary. This migration is followed by a fusion of the migrant and stationary micronuclei; this fusion effects a complete interchange of hereditary substance, after which the two Infusoria separate and enter upon a new life cycle. Meanwhile the meganucleus breaks up and is reconstituted in each fertilized cell.

Maupas gathers from these interesting phenomena additional proof that the chromatin of all cells bears the inherited characteristics and that the cytoplasm and nucleoplasm, or achromatin, is the dynamic agent, because the micronuclei bearing the chromatin are the only structures which are permanent and persistent, all the other structures—nucleoplasm, archoplasm, etc.—being replaced and renewed. The reduction of the chromatin is purely quantitative, the eliminated and fertilizing micronuclei being exactly equivalent; after the chromatin has been quartered the cell becomes incapable of further activity until it is reinforced by chromatin from the copulating cell.

No Distinction Between the Sexes in Heredity.—The three laws which underlie these phenomena are: 1. That fertilization consists in the union of the hereditary substance of two individuals. 2. That before union the hereditary substance in

each is greatly reduced. 3. That there is no line between male and female, the conjugating cells are simply in a similar physiological condition wherein a mingling of hereditary characteristics affords a new lease of life. As Maupas says:

"Les différences appelées sexuelles portent sur des faits et des phénomènes purement accessoires de la fécondation. La fécondation consiste uniquement dans la réunion et la copulation de deux noyaux semblables et équivalents, mais provenus de deux cellules distinctes."

In this conclusion as to the secondary and superficial, rather than fundamental, difference between the two sexes, Maupas simply confirms the views of Strasburger, the botanist, Hensen, R. and O. Hertwig, Weismann, and others, namely, that sex has evolved from the necessity of cell conjugation; that even in the higher forms the cells borne by the two sexes are absolutely neutral so far as sex is concerned, the wide difference of form of the germ-cells is a result of physiological division of labor—the mass and yolk of the ovum having been differentiated to support the early stages of development, while the spermatozoon has dispensed with all these accessories and acquired an active vibratile form for its function of reaching and penetrating the ovum. The evidence of the Infusoria is paralleled among some of the plants, in which conjugation between entirely similar cells is observed.

The causes finally determining sex may come surprisingly late in development, and according to the investigations of Düsing and the experiments of Yung[5] and of Giron are directly related to nutrition. High feeding favors an increase of the percentage of females, while, conversely, low feeding increases the males. In Yung's experiments with tadpoles the following results were obtained:

	Females.	Males.
Normal percentage	57	43
High nutrition	92	8

[5]See Geddes and Thomson: The Evolution of Sex, 1891, also, Düsing: Die Regulierung des Geschlechtsverhältnisses bei d. Vermehrung der Menschen, Tiere und Pflanzen, Jen. Zeit. f. Natur., Bd. 17, 1884.

Geddes expresses this principle in physiological terms of metabolism, that anabolic (constructive) conditions produce females, while katabolic (destructive) conditions produce males.

I think we may now safely eliminate the factor of sex from our calculations upon the problem of heredity, and thus rid ourselves of one of the oldest and most widespread fallacies. We shall thus, in using the terms "paternal" and "maternal" imply merely the distinction between two lines of family descent.

The Theory of Reduction.—This leads us back to the significance of the polar bodies. Van Beneden's discovery that these

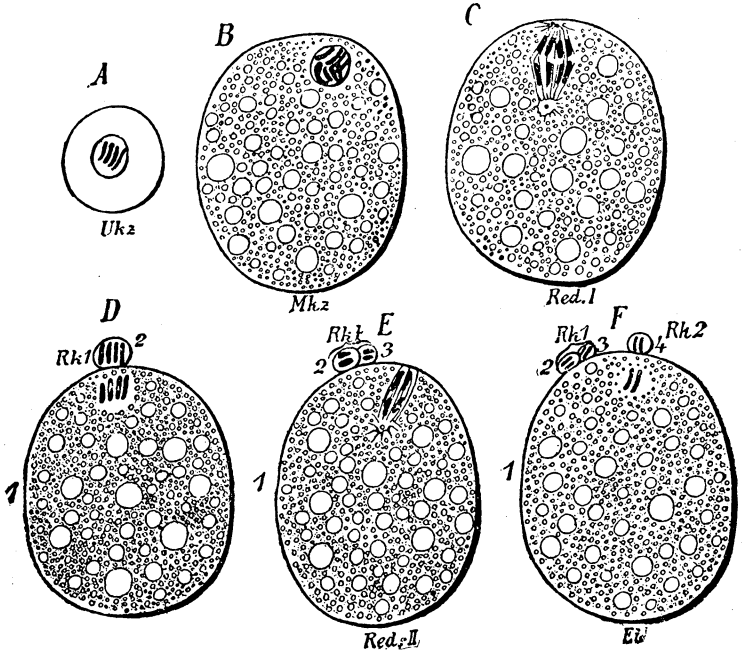

FIG. 11.—THE MATURATION OF OVA, OR FORMATION OF POLAR BODIES IN ASCARIS. (From Weismann after Hertwig.) A, original germ-cell in embryonic germ-layer—4 chromatin rods; B, Ovum mother-cell—8 rods; C-D, First polar body extruded; E, Splitting of first polar body. Ovum still contains 4 rods; F, Second polar body extruded; Ovum mature with 2 rods.

bodies contained chromatin led gradually to the view that they were not fragments of the ova, but represented minute morphologically complete cells. Bütschli showed that they were

given off independently of, and prior to, the contact of the spermatozoon, and, finding in the leeches that the first polar body subdivides to form two bodies, he considered them as formed by true cell division, and containing both nucleoplasm and chromatin. Giard independently reached a similar opinion, assigning an atavistic meaning to the polar cells. Whitman, in 1878, advanced the idea that they represented vestiges of the primitive mode of reproduction by fission, while Mark described them as "abortive ova."

At this point speculation subsided until it was revived by Weismann's attempt to connect these bodies with his theory of heredity,[6] already referred to. The whole history is clearly given in R. Hertwig's masterly memoir upon Ovo and Spermatogenesis in the Nematodes.[7] Taking advantage of Boveri's discoveries in staining technique, and stimulated by Weismann's prediction that spermatozoa would also be found to extrude polar bodies, this author examined all stages in the peculiarly favorable germ-cells of the thread-worm of the horse (*Ascaris megalocephala*).

He made the surprising discovery that ova and spermatozoa are formed in a substantially similar manner by repeated divisions, the single difference being that the last products of division among the sperm-cells are effective spermatozoa, capable of development in fertilization, while the last products of division in the ovary are, first, the true ova, and second, the abortive ova (polar cells) incapable of development. In both ova and spermatozoa the nucleus contains but one-half the chromatin which a typical nucleus contains; in the case of *A. megalocephala* each of the germ-cells contains but two chromasomes while the normal body-cells contain four. The manner in which this maturation of the germ-cells for conjugation is brought about is beautifully shown in these diagrams, taken from Weismann's essay, "Amphimixis." You observe that the number of chromasomes in the primary germ-cells is four (figs. 11 and 12, *A*). Then are formed by subdivision the ovum and sperm "mother-cells," in which the chromatin substance

[6]On the Number of Polar Bodies and their Significance in Heredity. 1887.

[7]Ei- und Samenbildung bei Nematoden, Archi. mikr. Anat., Bd. 26, 1890.

is doubled, so that we observe eight chromasomes. The mother-cells then divide and the chromatin is reduced to four rods, a second division rapidly follows whereby the chromatin is reduced to two rods, or half the original quantity. These last divisions take place by karyokinesis, but, as Hertwig points out, they differ from typical karyokinesis in the fact that the divisions follow so rapidly upon each other that the vesicular

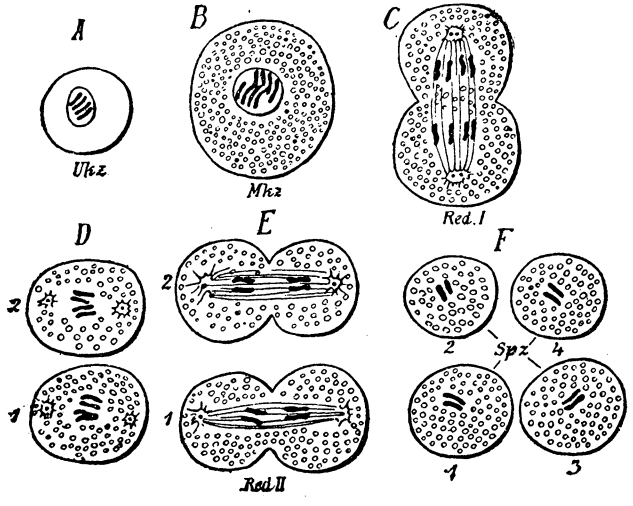

FIG. 12.--SPERMATOGENESIS IN ASCARIS. (From Weismann after Hertwig.) A, Original germ-cell—4 chromatin rods; B, Sperm mother-cell—8 rods; C-D, First daughter-cells with 4 rods each; E-F, Formation of second daughter-cells, or mature spermatozoa, with 2 rods each.

resting-period of the nucleus is omitted. Thus, he suggests, is prevented an over-accumulation of chromatin substance prior to the fusion of the ovum and sperm.

It is evident that the polar-cells are rudimentary ova, which do not possess the yolk-mass, etc., essential to development, and are divided off at a very late stage, sometimes after the egg has left the ovary, but are in other respects analogous to the spermatozoa. The reason these polar-cells have not disappeared altogether in either plants or animals is that they originally possessed a deep physiological importance. As the first polar-cell subdivides and forms two, it follows that from both ovum and sperm mother-cells four daughter-cells are

formed, each containing half the chromatin substance of a normal nucleus. In the ovary three of these daughter-cells abort and the fourth forms a true ovum; in the sperm-gland, however, all four daughter-cells form spermatozoa.

We may thus consider the polar-cell problem as in all probability settled; the whole process is probably an inheritance or survival of a primitive condition in which all four ova, like the four spermatozoa, were fully functional.

The Relation between the Chromatin and Heredity.—We have just seen that the last stages in the preparation of the ova and spermatozoa for conjugation result in halving the number of rods in the original germ-cells. Now, as Hertwig and Weismann point out, one point is still left in doubt. Why is the chromatin substance doubled in the mother-cells so that two successive subdivisions are necessary to reduce it to half the original quantity? Hertwig has not attempted to answer this question, as he prefers to wait for further research. Weismann, however, who is unfortunately cut off from research by failing eyesight, has offered a speculative solution to this problem which he trusts may guide future investigation.

This leads me to say a few words in regard to his conception of the relation of the chromatin to heredity. 1. His first premise is that in fertilization there is not a fusion of chromatin but that a certain independence is preserved between the maternal and paternal elements, based upon the observed fact that the two pairs of rods do not fuse but lie side by side, and upon the assumption that these pairs are kept distinct in each cell through all the subsequent stages of embryonic and adult development. If this be the case, the hereditary substance contributed by the father would remain separate from that contributed by the mother, throughout. 2. "Each of these pairs would be made up of the collective predispositions which are indispensable for the building up of an individual, but each possesses an individual character, for they are not entirely alike. I have called such units "ancestral plasms," and I conceive that they are contained in numbers in the chromatin of the mature germ-cells of living organisms, also that the older nuclear rods are made up of a certain number of these.

. . . Obviously these units cannot become infinitely minute; however small they may be they must always retain a certain size. This follows from the extremely complicated structure which we must without any doubt ascribe to them." These units are not, however, ultimate, they are in turn extremely complex, and are composed of countless biological units of the kind conceived by Nägeli and others. 3. The reduction of the chromatin only acquires a meaning when taken in connection with the above supposition of distinct ancestral plasms, and has no meaning if we accept Hertwig's view that there is a complete fusion of maternal and paternal germ-plasm. This meaning is that reduction in the maturation of germ-cells is *sui generis*, it does not divide the ancestral plasms into two similar groups, but one daughter-cell receives one set of germ-plasms or hereditary predispositions, and another daughter-cell receives another; reduction is thus differential. According to this view the four sperm and ovum daughter-cells would each contain a different set of ancestral plasms. 4. The fact that the chromatin substance is doubled in the sperm and ovum mother-cells, so that we observe double the number of rods characteristic of the species, is to be explained as an adaptation to the requirements of natural selection, for this doubling and subsequent double division render possible an infinite number of combinations (as many, in fact, as there are individuals) for Selection to operate upon.

This explanation of Weismann's is an example of his apotheosis of the theory of natural selection. Every process is made to suit this theory, which, as we have seen in the first and second lectures, is, in his opinion, the exclusive factor of evolution. But this very high degree of mingling and remingling of ancestral predispositions would be fatal to evolution, for after a combination favorable to survival had been established in one generation it would be broken up into a new combination, perhaps unfavorable to survival, in the next generation. This entire essay upon "Amphimixis," or the theory of mingling of reduced hereditary substance, will, I believe, mark a turning-point to decline in Weismann's influence as a biolo-

gist. His whole reasoning is now in a circle around the natural selection theory.

The Meaning of Conjugation.—Weismann looks upon sexual reproduction as designed to mingle hereditary tendencies and to create individual differences whereby natural selection may form new species. It is evident that these combinations must be mainly fortuitous and productive of indefinite variation; but we have seen that evolution advances largely by the accumulation of definite variations, or those in which each successive generation exhibits the same tendencies to depart from the typical ancestral form in certain parts of the body, and that these tendencies stand out in relief among the diffused kaleidoscopic or fortuitous anomalies.

The fact, moreover, that variability and evolution by the accumulation of certain variations in successive generations is also observed in organisms which reproduce asexually, both among plants and animals, shows that we must look in another direction for the underlying cause or purpose of sexual reproduction. Weismann rightly combats the old idea of "vitalization" of the ovum by the spermatozoon, and it is perfectly evident from the researches of Maupas and Hertwig that the ovum may as accurately be said to vitalize the spermatazoon as the reverse. Fecundation is simply the approximation of two hereditary substances of distinct origin and their incorporation into a single nucleus. The action and reaction of these substances may be considered equal and mutual so far as we now know.

The remarkably ingenious experiments of Hertwig and Boveri, above alluded to, strengthen this idea. Some years ago Weismann wrote: "If it were possible to introduce the female pronucleus of an egg into another egg of the same species, immediately after the transformation of the nucleus of the latter into the female pronucleus, it is very probable that the two nuclei would conjugate just as if a fertilizing sperm-nucleus had penetrated. If this were so, the direct proof that egg-nucleus and sperm-nucleus are identical would be furnished." Boveri succeeded in accomplishing a similar feat by depriving an ovum of its nucleus and subsequently causing it

to develop by admitting *a spermatozoan which fertilized the denucleated ovum and produced a complete individual!*

In opposing the vitalizing properties of the sperm, Weismann, however, went further, and advocated the view that there is nothing in the nature of vitalization or "rejuvenescence" in conjugation—that, given proper environment, protoplasm is immortal, and runs upon a course of undiminished activity. This we have seen is not the case in the Infusoria, and, as recently remarked by Hartog, there is only one class of organisms which, according to our present knowledge, are completely agamous and immortal—namely, the Monadina. It may in future appear that even in the monads there is a cycle for the development in which conjugation plays its part.

Maupas's experiments seem to establish the primitive, and therefore the true, interpretation of the purpose of conjugation as well as of sex, the latter being a consequence of the former, namely, that after a long period of direct subdivision of hereditary material from a single individual, a limit is reached beyond which the forces of heredity are not reproduced in their original intensity unless combined with another set of similar forces of different origin. This combination restores the original intensity. It is objected to this that two sets of feeble forces cannot constitute one vigorous force, but this is met by the observed fact that such union does start a new life cycle, and is therefore rejuvenescent. We may regard this as the fundamental meaning of conjugation and the production of variations as entirely secondary.

The Distribution of the Chromatin.—We have now reviewed some of the main phenomena of fertilization; there still remains the relation of the hereditary substance to the future development of the individual. There is, first, the astonishing fact that, as the chromatin goes on dividing, its mass or volume remains apparently undiminished; that is, there is apparently as much chromatin in one of the many million active cells of the body as in the original fertilized ovum, and there is still an enigma as to the nature of this chromatin and its functions. Secondly, there is the problem of the maternal and

paternal elements in each cell; do they lie side by side or are they fused?

1st. In plants De Vries[8] and others believe that all or by far the greater number of cells in the plant body contain the total hereditary characters of the species in a latent condition. Kölliker[9] has fully discussed this question and called attention to Müller's early views that, in spite of the physiological division of labor producing the tissues, the properties of all the tissues can be derived from the nuclear substance of a single tissue, as proved by experiments upon the lower animals. Weismann, on the other hand, has held that the course of development is marked by a constant qualitative distribution of his germ-plasm or hereditary substance, so that, so far as nuclear content is concerned, there are three forms of cells: 1, with nucleoplasm; 2, with nucleoplasm and germ-plasm; 3, with germ-plasm only. Kölliker opposes this idea and maintains that the "idioplasma" passes into all cells, in which it divides in course of development; step by step from the embryonic layers to the tissues, the constructive processes are under the direction of the nuclei containing this hereditary substance; it remains in every nucleus for a long period unaltered, in order to finally, here earlier, there later, impress its constructive forces. In certain elements, as in blood-corpuscles, epidermal scales, etc., it disappears, as the last product of division.

R. Hertwig takes a similar view; since embryonic and adult cell division is differential there must be a form of differentiation in the nucleus, but this does not consist in the total elimination of some qualities and survival of others, nor of a reduction in mass. The mass and the properties remain the same in every cell, the differentiation consists in the activity of certain elements in certain tissues. Thus we may say with De Vries, that different "pangene" may leave the nucleus and enter the cell in different tissues, or with Nägeli, that special "micellæ" come into activity at certain points; in other words,

[8]Hugo de Vries: Intracellulare Pangenesis. Jena, 1889.

[9]Die Bedeutung der Zellkerne für die Vorgänge der Vererbung, Zeit. f. wiss. Zoöl., 1885. And, Das Karyoplasma und die Vererbung, op. cit., 1886.

the potential of the nucleus is differently exerted. Here, again, we have the idea of patent and latent hereditary elements, such as appear in the entire individual upon a larger scale.

This is one of the most interesting problems for future investigation, but the direction of research will, I imagine, cover a larger area of cell-content than the nucleus, as we are now swinging back to regard the extra nuclear archoplasm as an important factor in the process.

In the following paragraph Hertwig expressed his view of nuclear control and cytoplasmic differentiation:

"As I saw in the transformation of the nucleus during fertilization proof that it is the bearer of hereditary substance I recognized a great advance in the fact that the nucleus leaves in the same form in every cell, and in its vesicular capsule is somewhat removed from the metamorphoses of the cells. As Nägeli spread his idioplasm as a net-work throughout the whole body, so, according to my theory, every body-cell contained in its nucleus its quota of hereditary substance, while its specific histological peculiarities were to be regarded as its plasma-products."

2d. The next question *is the fate of the maternal and paternal contributions to the embryo.* Here there is a wide difference of opinion. On the one side Van Beneden is the leader of those who regard each cell of the body as in a sense hermaphrodite; as we have seen, his views of maturation and the significance of the extension of the polar bodies were colored by this theory, for he regarded the germ-cells as hermaphrodite until one sex was eliminated. But now that the researches of Hertwig have given the last blow to Van Beneden's theory, and it follows that there can be no male and female chromasomes, there still remains room for the analogous view that the maternal and paternal chromasomes remain distinct throughout the course of development, not as sexual elements but as substances with the same racial and specific but different individual tendencies. Rabl, an eminent embryologist, shares this view, and it is supported by Boveri upon the observation that in each division the paternal and maternal elements are kept distinct, and in *Ascaris*, for example, two of the chromasomes

of each division figure are paternal and two are maternal. In favor of this hypothesis we may place the following facts: 1st, that there are an even number of chromasome rods in all cells; 2d, that the number is constant throughout all the subsequent changes in the tissues; 3d, that the number is fixed for each species or variety; 4th, that the number is the same in each sex.

Against this *replacement* hypothesis we must consider the extreme complexity of the division process, and the long-resting, or thread stage, in which the chromatin lies in a confused coil. Further, Hertwig argues that if the elements are distinct we should find some evidence that the maternal or paternal part is atrophied or replaced, or excluded from the nucleus, for both parts cannot share alike in the control of the cell. These are Hertwig's grounds for supporting the "verschmelsungstheorie," or *fusion* theory, also advocated by Waldeyer, to the effect that by the complete union of the maternal and paternal substance a new product is formed; in this fusion the law of prepotency may come into play, causing one or other of the parental tendencies to predominate, or there may be an even redistribution, whereby, as expressed by Hensen, "the hereditary substance of the son is not that of the father plus that of the mother, but is his own, with a new hereditary form resulting from the combination."

While suspending judgment between these two views as to the separation or fusion of the chromatin, we may appeal to the external phenomena of heredity for light upon the probabilities in the question. First, I refer to the very decided opinion of Francis Galton in regard to particulate inheritance; he is so impressed with the fact that we are made up bit by bit of separate structures derived from different ancestors that he has even suggested that the skin of the mulatto may represent not a fusion of white and black but an excessively fine mosaic in which the colors are so distributed as to give the appearance of blending. We do sometimes observe patches of color as evidence of uneven distribution. As Galton distinguishes two types of structures with reference to inheritance, viz., those which blend and those which do not blend, we might corre-

late these types with prepotency, replacement, and fusion. Where characteristics do not blend, as in eye-color, it is evident that, while the offspring must receive from both parents the material basis for the formation of the complete color of the eye, either the maternal or paternal material must be prepotent and exclude the development of the other; the logical inference is that the former actively replaces the latter; but it is not necessary that exclusion from the cell chromatin should follow. Now, while some blends seem to support the theory of fusion, the sum total of facts of heredity are strongly against this as a universal principle, for many maternal and paternal structures are preserved in their absolute integrity for generations without the least indication of mixture.

Cell Forces and Heredity.—We have thus far been considering only the chromatin as the heredity substance *par excellence*, and have disregarded for the time the archoplasm or dynamic material of the cell. If we advance upon the hypothesis that a typical cell contains the more or less passive chromatin, and the archoplasm playing upon this chromatin in course of every phase of redistribution, it seems *á priori* improbable that elements which are associated with every vital change should be dissociated in the phenomena of heredity. We might suppose that the mechanics of karyokinesis are exactly similar in every cell of one individual, but it is highly improbable that they should be exactly similar in two individuals. We should, therefore anticipate the joint transmission of the chromatin and archoplasm, implying by the latter the dynamic centers especially connected with hereditary function as distinguished from the general functions of metabolism.

This leads us to look for evidence from the life of the cell in its totality. We owe to Dr. Max Verworn[10] a fresh treatment of this subject, based upon experimental researches among the Infusoria, mainly by the extirpation method. As his experiments included only the phenomena of living cells

[10]Die Physiologische Bedeutung des Zellkerns, Archiv für Physiologie, 1891, pp. 113–115.

in which the chromatin substance was of course undifferentiated to the eye, he treats of the nucleus as a whole without distinction as to chromatin and achromatin. He concludes that the physiological importance of the nucleus is exhibited in its constant interchange of materials with the remainder of the cell body, only through this interchange does it influence the cell and control its life processes. The interchange is in triple currents, *a*, from outside of cell to cytoplasm; *b*, from cytoplasm to nucleus; *c*, from nucleus to cytoplasm. These movements of interchange are the expression of life phenomena. He compares the *rôle* of the nucleus to that of a cell organoid, like chlorophyll, as not constantly present but as invariably necessary to activity. Thus he believes even the most lowly organized cells have nuclear centres, and that even bacteria are differentiated into nuclear and extra-nuclear areas. Coupled with this idea of nuclear control is the somewhat paradoxical statement that the nucleus is not a dynamic centre, either automatic or regulating, and the conclusion that the nucleus alone cannot be the seat of fertilization and heredity, but both the nucleus and extra-nuclear protoplasm must constitute the material basis of heredity. This conclusion is in the direction of the general reaction of opinion which is now taking place against the centralization of cell-government in the nucleus.

Vague as they must necessarily be, our ideas of cell forces are somewhat further defined by the brilliant experiments of the Hertwig brothers upon germ-cell physiology and pathology, which are full of suggestion as to the causation of abnormalities in inheritance. These were begun in 1884 and were first directed to the influence of gravitation upon the planes of embryonic cell division, following up the experiments of Pflüger and Rauber. In 1885 the conditions of bastard fertilization were studied; in 1887 the causes of polyspermy or multiple fertilization; and in 1890 the effects of extreme heat and cold upon germ-cell functions.[11] In general the conclusions reached were that in the normal state there exist regulating

[11] Experimentelle Untersuchungen über die Bedingungen der Bastardbefruchtung, Jena, 1885. See series of papers in Jenaische Zeitschrift.

forces in the ovum which prevent multiple fertilization or bastard fertilization (*i. e.*, by spermatozoa of other varieties), but these forces are neutralized where the life-energy of the cell is diminished by reagents or by extremes of temperature.

For example, in the normal state the entrance of a single spermatozoon produces a reaction in the ovum-wall preventing the entrance of other spermatozoa, but when the ovum is weakened by chloroform solution two or more spermatozoa enter before the reaction appears; in fact the degree of polyspermy is directly proportional to the intensity of the chemical, thermic, or mechanical disturbance of the ovum. Double fertilization or over-fertilization has not in a single case resulted in the production of twins, so that Fol's supposition is negatived, although other forms may behave differently. The cell-function may be arrested at any stage by thermic influences; thus two pronuclei, paternal and maternal, about to unite can be held apart by lowering the temperature. Polyspermy also results from a lower temperature. It is noteworthy that the conditions of bastard fertilization and polyspermy are different; chloroform produces the latter but not the former. Kupffer has, I believe, succeeded in producing twins, or rather two-headed monsters, by abnormal fertilization in fishes.

These researches, although made with a different object, re-establish the older views as to the interdependence of nuclear and extra nuclear activities, and show that no sharp line of demarcation of function can be drawn between the nucleus as a center of reproduction and heredity, and the cytoplasm as the seat of tissue-building and nutrition. In Boveri's discovery of the archoplasmic centres, or centrosomes, we find positive ground for this broader view. It is connected with the cell phenomena of heredity in the following manner:

While the union of the nuclei in fertilization is the most obvious feature, this union is dependent upon the archoplasm, which rearranges the nuclear elements. If the spermatozoon contains no archoplasm, this power cannot come from the paternal side; but Boveri shows that this is probably not the case and that the spermatozoon brings its centrosome with it, thus entering the ovum with both the paternal chromatin sub-

stance and dynamic material. It is certain from this and from the observations of Roux that the sperm-cell is now to be regarded as more than a mere nucleus, that it contains both nuclein and para-nuclein.

Intercellular Forces.—The forces within the different portions of the cell lead us to consider those which must exist between different cells. This is an obscure question at present; but, as I have observed in the close of the second lecture, it is an extremely important one in connection with the problem of heredity. As Professor Wilson writes, "My own conviction steadily grows that the cell is not a self-regulating mechanism in itself, that no cell is isolated, and that Weismann's fundamental proposition is false."

It is a long step between an *á priori* conviction and the demonstration by experiment of a correlation of forces between the cells. This seems to me a most important field of experiment. We have seen in Maupas's work that the contact of two Infusoria initiates a rapid series of internal changes; we have only to conceive of analogous changes taking place when two cells are not in actual contact, as in the phenomena of previous fertilization referred to in my second lecture. Hertwig and others have shown how gravitation is related to cell activity. Roux has destroyed half an embryo with a hot needle in the first stages of segmentation and followed the other half through the stages of subsequent development. Another clever experimenter has turned fertilized ova upside down during the early stages of development, and shown how the protoplasmic pole and yolk-pole forcibly change places. Driesch has traced the connection and meaning of the first plane of cleavage in the embryos of echinoderms, and has succeeded in raising a small adult from half an embryo artificially separated during the first cleavage stage. Wilson, in the larva of Nereis, has shown how a certain stage of division in one group of cells affects all the other groups. All these experiments are in the line of determining the relation which exists between internal cell forces and other natural forces. What we must now seek to determine is the relation of cell to cell throughout the body, in connection with the phenomena of heredity.

Conclusions.—The most impressive truth issuing from our review of recent researches in evolution and heredity is the uniformity of life-processes throughout the whole scale of life from the Infusoria to man. The most striking analogy is that seen in the laws of fertilization and conjugation, which are shown by Maupas's researches to have been established substantially in their present form at a very early period in the evolution of living organisms. Such uniformity furnishes a powerful argument for the advocates of the study of biology as an introduction to the applied science of medicine. Much that is now entirely omitted from medical education, because it is considered too remote, is in reality at the very roots of the science. To understand the disorders of life we should first thoroughly understand the essential phenomena of normal life. Of course we shall never see life as it really is, because there is always something beyond our highest magnifying powers; but we come nearest to this invisible form of energy when, with such investigators as Hertwig and Maupas, we strip the life-processes of all their accessories and view them in their simplest external form.

The problems of evolution are found to be inseparably connected with those of heredity. No theory is at all adequate which does not explain both classes of facts, and we have seen that the explanations offered by the two opposed schools—those who believe in the transmission of acquired characters and those who do not—are directly exclusive of each other. We should suspend judgment entirely rather than cease to gather from every quarter facts which bear upon the most important and central problem of the transmission of acquired characters. I have endeavored to point out the opportunities which medical practitioners enjoy of contributing evidence upon this mooted question. It must not be forgotten that while the inheritance of individual adaptation to environment is the simplest method of explaining race adaptation such as we observe in the evolution of man, we know absolutely nothing of how such inheritance can be effected through the germ-cells. We cannot at present construct even any form of working hypothesis for such a process. On the other hand, we

have found how untenable is the alternative theory offered to us by Weismann, that it is solely natural selection or the survival of the fittest which

> ". . . shapes our ends,
> Rough-hew them as we will."

At the same time Weismann's conception of a continuity of germinal protoplasm, which we have found to consist in chromatin plus archoplasm, helps us over many of the phenomena of heredity, especially on the retrogressive side, and if it were not that we must also account for progressive and definite transformation in heredity, we might credit the distinguished Freiburg naturalist with having loosed the Gordian knot.

In summing up, the order of treatment followed in the lectures may be reversed, and we can begin with the germ-cells, and condense the more or less ascertained facts.

The Germ-cells.—1. The material substance of hereditary transmission is the highly coloring protoplasm, or chromatin, in the nucleus of the germ-cells, probably connected with a certain form of archoplasm, or dynamic protoplasm, outside of the nucleus.

2. Before conjugation and fertilization the hereditary substance of both the male and female cells is reduced to one-half that found in a typical cell. The substance is, however, first doubled and then quartered, the meaning of which process is not understood.

3. There is a difference of opinion as to whether the paternal and maternal hereditary substances, are fused or lie side by side during fertilization, also as to how the substance is distributed through the tissues, during individual growth, whether *en masse* or by qualitative distribution.

Heredity.—4. No form of physical connection between the germ-cells and body-cells is known, but the facts of heredity seem to render such a connection theoretically necessary. Several classes of facts witnessed in reproduction seem to support this theory.

5. The facts of Heredity support the theory of a continuous and specific form of protoplasm as the basis of repetition of type.

Evolution.—6. The facts of evolution, both in present and past time, point to transformism by definite progression toward new types of structure in succeeding generations, opposing the retrogressive forces of heredity.

7. The theory (Natural Selection) of definite progression by the accumulation of fortuitous favorable variations is found to be not only theoretically improbable, but not to correspond with the observed laws of variation.

8. The laws of variation (anomalies) lend support to the theory of hereditary transmission of individual acquired variations, but even this (Lamarckian) theory encounters many difficulties.

I think this is as fair a statement as can be made at the present time, and it rests upon a general survey of the whole field.